

INTERNATIONAL STANDARD

ISO 22612

First edition
2005-03-01

Clothing for protection against infectious agents — Test method for resistance to dry microbial penetration

*Vêtements de protection contre les agents infectieux — Méthode
d'essai de la résistance à la pénétration microbienne par voie sèche*



Reference number
ISO 22612:2005(E)

© ISO 2005

PDF disclaimer

This PDF file may contain embedded typefaces. In accordance with Adobe's licensing policy, this file may be printed or viewed but shall not be edited unless the typefaces which are embedded are licensed to and installed on the computer performing the editing. In downloading this file, parties accept therein the responsibility of not infringing Adobe's licensing policy. The ISO Central Secretariat accepts no liability in this area.

Adobe is a trademark of Adobe Systems Incorporated.

Details of the software products used to create this PDF file can be found in the General Info relative to the file; the PDF-creation parameters were optimized for printing. Every care has been taken to ensure that the file is suitable for use by ISO member bodies. In the unlikely event that a problem relating to it is found, please inform the Central Secretariat at the address given below.

© ISO 2005

All rights reserved. Unless otherwise specified, no part of this publication may be reproduced or utilized in any form or by any means, electronic or mechanical, including photocopying and microfilm, without permission in writing from either ISO at the address below or ISO's member body in the country of the requester.

ISO copyright office
Case postale 56 • CH-1211 Geneva 20
Tel. + 41 22 749 01 11
Fax + 41 22 749 09 47
E-mail copyright@iso.org
Web www.iso.org

Published in Switzerland

Foreword

ISO (the International Organization for Standardization) is a worldwide federation of national standards bodies (ISO member bodies). The work of preparing International Standards is normally carried out through ISO technical committees. Each member body interested in a subject for which a technical committee has been established has the right to be represented on that committee. International organizations, governmental and non-governmental, in liaison with ISO, also take part in the work. ISO collaborates closely with the International Electrotechnical Commission (IEC) on all matters of electrotechnical standardization.

International Standards are drafted in accordance with the rules given in the ISO/IEC Directives, Part 2.

The main task of technical committees is to prepare International Standards. Draft International Standards adopted by the technical committees are circulated to the member bodies for voting. Publication as an International Standard requires approval by at least 75 % of the member bodies casting a vote.

Attention is drawn to the possibility that some of the elements of this document may be the subject of patent rights. ISO shall not be held responsible for identifying any or all such patent rights.

ISO 22612 was prepared by the European Committee for Standardization (CEN) in collaboration with Technical Committee ISO/TC 94, *Personal safety — Protective clothing and equipment*, Subcommittee SC 13, *Protective clothing*, in accordance with the Agreement on technical cooperation between ISO and CEN (Vienna Agreement).

Throughout the text of this document, read “...this European Standard...” to mean “...this International Standard...”.

For the purposes of this International Standard, the CEN annexes regarding the fulfilment of the European Council Directives have been removed.

Contents

| | page |
|--|------|
| Foreword..... | v |
| Introduction | vi |
| 1 Scope | 1 |
| 2 Normative references | 1 |
| 3 Terms and definitions | 1 |
| 4 Principle | 1 |
| 5 Testing conditions | 1 |
| 6 Equipment | 1 |
| 6.1 General lay-out..... | 1 |
| 6.2 Test containers | 2 |
| 6.3 Method to infect talc with spores..... | 2 |
| 6.3.1 Materials | 2 |
| 6.3.2 Procedure | 3 |
| 7 Procedure | 3 |
| 8 Test report | 4 |
| Annex A (informative) Preparation of TGE agar medium | 7 |
| A.1 Ingredients..... | 7 |
| A.2 Procedure | 7 |
| Bibliography..... | 8 |

Foreword

This document (EN ISO 22612:2005) has been prepared by Technical Committee CEN/TC 205 “Non-active medical devices”, the secretariat of which is held by BSI, in collaboration with Technical Committee ISO/TC 94 “Personal safety - Protective clothing and equipment”.

This European Standard shall be given the status of a national standard, either by publication of an identical text or by endorsement, at the latest by August 2005, and conflicting national standards shall be withdrawn at the latest by August 2005.

This document has been prepared under a mandate given to CEN by the European Commission and the European Free Trade Association, and supports essential requirements of EU Directive(s).

According to the CEN/CENELEC Internal Regulations, the national standards organizations of the following countries are bound to implement this European Standard: Austria, Belgium, Cyprus, Czech Republic, Denmark, Estonia, Finland, France, Germany, Greece, Hungary, Iceland, Ireland, Italy, Latvia, Lithuania, Luxembourg, Malta, Netherlands, Norway, Poland, Portugal, Slovakia, Slovenia, Spain, Sweden, Switzerland and United Kingdom.

Introduction

There are numerous examples of situations where bacteria may migrate through a barrier material in the dry state carried by organic or inorganic particles. The dry penetration of bacteria-carrying skin scales through an operating gown or a clean air suit is one example. Penetration through a packaging material during storage is another.

This document EN ISO 22612 describes a test method, with the associated equipment, that may be used to determine a material's resistance to dry penetration of bacteria on particles in the size range most typical for human skin scales

1 Scope

This test method provides a means for assessing the resistance to penetration through barrier materials of bacteria-carrying particles.

NOTE Due to its complexity, this EN ISO 22612 cannot be considered as a useful method for routine quality control but may suit the needs when a material is assessed for compliance with the requirements of current regulations such as EU Directive 93/42/EEC.

2 Normative references

The following referenced document is indispensable for the application of this document. For dated references, only the edition cited applies. For undated references, the latest edition of the referenced document (including any amendments) applies.

EN 13795-1:2002, *Surgical drapes, gowns and clean air suits, used as medical devices for patients, clinical staff and equipment – Part 1: General requirements for manufacturers, processors and products.*

3 Terms and definitions

For the purposes of this document, the terms and definitions given in EN 13795-1:2002 apply.

4 Principle

The test is carried out on test pieces each fixed in a container. In every container except one a portion of talc contaminated with *Bacillus subtilis* is poured on the test piece. One container is left uncontaminated as a control. A sedimentation plate is inserted at the base of each container at a short distance below the test piece.

The apparatus supporting the containers is then vibrated by a pneumatic ball vibrator. The talc that penetrates is captured on the sedimentation plate. The sedimentation plates are removed and incubated.

The numbers of colonies produced are counted.

This document specifies two levels of challenge by means of giving two concentrations of bacterial cells on the talc particles and two times during which the barrier is subjected to vibration. The conditions for testing differ among product types and will be specified in other standards where this test method is applied such as in prEN 13795-3.

5 Testing conditions

Condition the samples and test at (20 ± 2) °C and (65 ± 5) % relative humidity.

6 Equipment

6.1 General lay-out

NOTE See Figure 1.

ISO 22612:2005(E)

6.1.1 A 10 mm thick stone plate, such as marble, 40 cm x 40 cm, underneath which 4 rubber stoppers are mounted at the corners.

6.1.2 A pneumatic ball vibrator¹⁾, able to generate 20 800 vibrations per minute with a force of 650 N.

6.1.3 The vibrator is attached by means of screws to the upper surface of the marble plate along one of its sides.

6.1.4 A compressed air flow meter capable of measuring the flow of air required to achieve a vibration frequency of 20 800 vibrations per minute.

6.1.5 Six stainless steel test containers.

6.1.6 A stainless steel plate with 6 retaining holes of suitable dimensions to fit the containers, the plate being held to the stone plate by means of clips.

6.1.7 Stopwatch.

6.2 Test containers

NOTE See Figure 2.

6.2.1 A suitable stainless steel container with a lid. The lid has a central aperture through which a metal plunger may be inserted to reach 10 mm underneath the lid to ensure that the test material is slack when inserted.

6.2.2 Each container has a sedimentation plate insertion slot near the base.

6.2.3 To ensure good contact between the containers and the stone plate by means of the fixing plate, each container is equipped with a rubber ring resting on its flanged base.

6.2.4 The rim of the container is chamfered to prevent damage to the test piece when inserted.

6.2.5 A supply of 9 cm diameter Petri dishes containing TGE agar (see Annex A).

6.3 Method to infect talc with spores

6.3.1 Materials

6.3.1.1 50 g ± 0,5 g of talc (95 % < 15 μm)²⁾

6.3.1.2 Purified spores of *Bacillus subtilis* ATCC 9372 at a concentration of ≥ 10⁹ /ml of ethyl alcohol ³⁾.

6.3.1.3 TGE agar plates.

1) e.g. K13, made by ERKALAITE OY, Helsinki, Finland. This information is given for the convenience of users of this document and does not constitute an endorsement by CEN of this product.

2) e.g. FINNTALC M15 from OMYA BENELUX S.A., Place Eug. Keym 43 B 27, B-1170, Bruxelles, tel.: +32 26 74 23 11, fax. +32 2672 92 68. This information is given for the convenience of users of this document and does not constitute an endorsement by CEN of this product.

3) e.g. SIMICON GmbH, Schuhmacherring 12, D-81737 München, fax +49 89 67 33 66 22. This information is given for the convenience of users of this document and does not constitute an endorsement by CEN of this product.

6.3.2 Procedure

- 6.3.2.1** Prepare 50 g sterile talc in a suitable container and sterilise at 160°C with dry heat for (2 – 0/+ 1) h.
- 6.3.2.2** Open an ampoule of 5 ml of the ethanolic spore solution.
- 6.3.2.3** Spread the spore solution in 50 steps (50 X 100 μ l) over the talc.
- 6.3.2.4** After every step shake the closed vessel with a vortex vibrator.
- 6.3.2.5** Put the opened vessel in a desiccator with silica gel and dry it at room temperature for 2 days to 3 days.
- 6.3.2.6** Weigh the vessel before and after drying to ensure complete drying.
- 6.3.2.7** Estimate the bioburden expressed as cfu/g (3 fold, each fold two times repeated) of the spore talc mixture on TGE agar after incubation overnight at 35 °C.
- 6.3.2.8** The final concentration should be 10⁴ or 10⁸ cfu/g talc. Ensure that the spores are homogeneously distributed in the talc.

7 Procedure

- 7.1** Cut 12 test pieces 200 mm x 200 mm.
- 7.2** Put test pieces in sterilising bags and sterilize by the method given by the manufacturer.
- 7.3** Put containers in sterilising bags and sterilize.
- 7.4** Fix the bases of the containers onto the stone plate by means of the fixing plate and secure with the clips.
- 7.5** Aseptically remove the pieces of test material from the bags and place over the mouths of the test containers.
- 7.6** With the plungers distended downwards, affix the lids to the containers thus fixing the test pieces with controlled slackness.
- 7.7** Remove the plungers.
- 7.8** Pour a 0,5 g \pm 0,1 g portion of contaminated talc through each plunger orifice onto 5 of the test materials leaving the 6th one uncontaminated as a control.
- 7.9** Seal the orifices with cling film.
- 7.10** Put a small plastic bag over each container.
- 7.11** A lidless sedimentation plate is inserted through the slot at base of each container.
- 7.12** Close the slots with adhesive tape.
- 7.13** Run the vibrator at an air flow that achieves vibration frequency of 20 800 vibrations per minute.
- 7.14** Remove plastic bags and adhesive tape.
- 7.15** Insert the lids of the sedimentation plates through the slots.

7.16 Remove sedimentation plates and incubate at 35 °C for 24 h.

7.17 Count the number of colonies produced. The control plate (6th) should read 0. If not the test should be aborted as there is extraneous contamination.

7.18 For each material repeat steps 7.1 to 7.17.

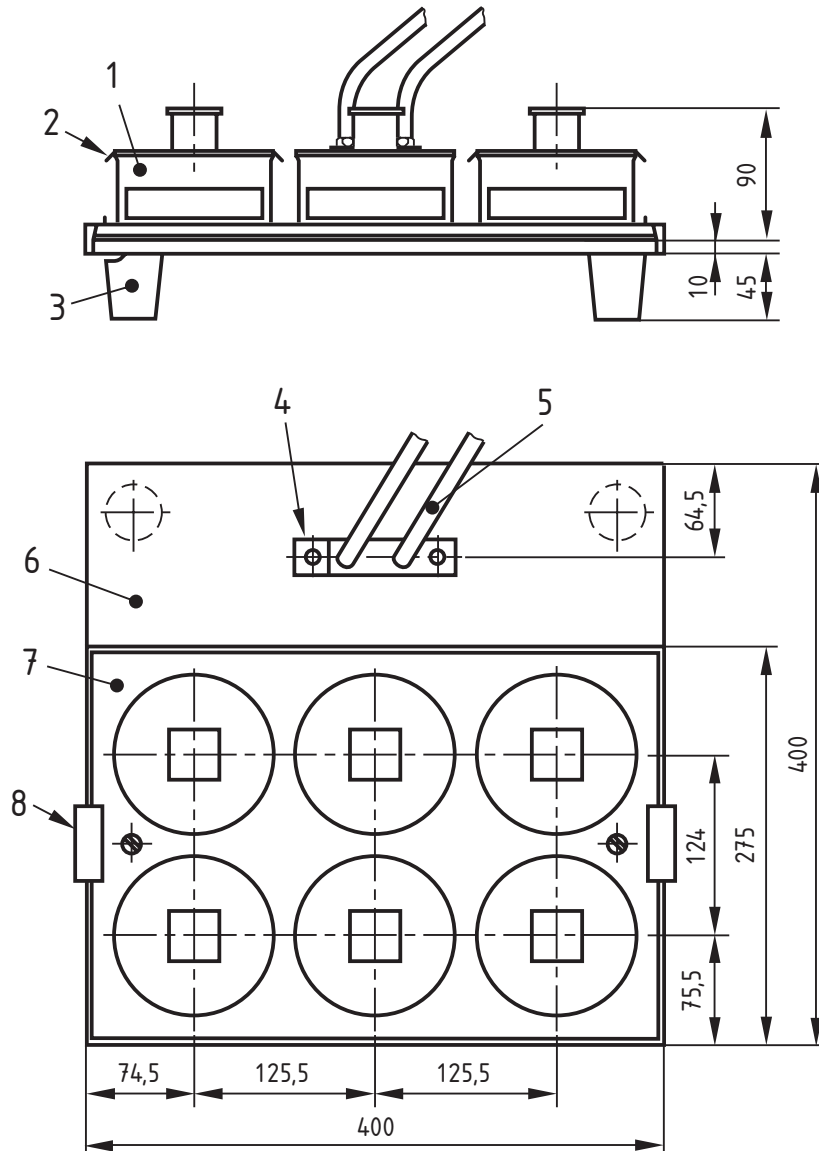
7.19 Calculate the arithmetic mean for the 10 valid results.

8 Test report

The test report shall include the following information:

- a) identity of material tested;
- b) test conditions, especially those variations chosen in 6.3.2.8 and 7.13;
- c) number of test pieces tested;
- d) any deviations from the standard method;
- e) details of the contaminant used;
- f) geometric mean of bacterial count (see 7.19).

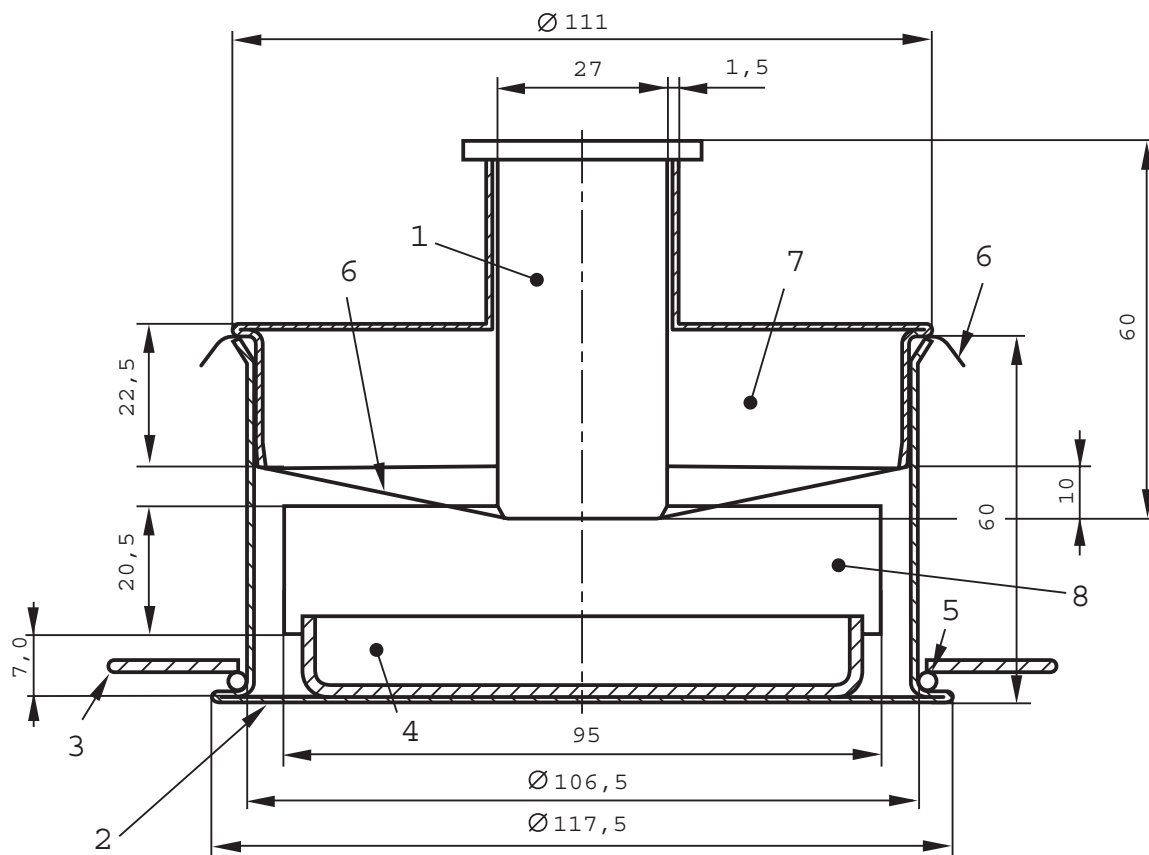
Dimensions in millimetres



Key

- 1 Test container
- 2 Test material
- 3 Rubber stoppers
- 4 Pneumatic ball vibrator
- 5 Pneumatic hoses
- 6 Marble plate
- 7 Fixing plate
- 8 Clips

Figure 1 — General layout of test equipment



Key

- 1 Metal plunger
- 2 Base of container
- 3 Fixing plate
- 4 Sedimentation plate
- 5 Rubber ring
- 6 Test material
- 7 Lid
- 8 Sedimentation plate insertion slot

Figure 2 — Test container

Annex A (informative)

Preparation of TGE agar medium

A.1 Ingredients

| | |
|--------------------|----------|
| Beef extract | 3 g |
| Tryptone | 5 g |
| Dextrose (glucose) | 1 g |
| Agar | 15 g |
| Distilled water | 1 000 ml |

A.2 Procedure

The solid ingredients are dissolved completely in the boiling distilled water and the pH is adjusted if necessary using dilute solutions of hydrochloric acid or sodium hydroxide so that the pH of the cooled agar at 25 °C will be $7,0 \pm 0,2$. It is sterilized by steam for 15 min at 120 °C. When appropriately cooled, plates are poured.

Bibliography

- [1] prEN 13795-3⁴, *Surgical drapes, gowns and clean air suits, used as medical devices, for patients, clinical staff and equipment – Part 3: Performance requirements and performance levels.*

⁴ To be published.

ISO 15926-2:2014

ICS 13.340.10

Price based on 8 pages